



PA (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
 XX  
 PI Newton DL, Rybak SM;  
 XX  
 DR MPI: 1999-610847/52.  
 DR N-PSDB: AA208132.  
 XX  
 PT New recombinant ribonucleases, used for killing target cells, e.g. for  
 PT treating cancers, viral infections or autoimmune diseases -  
 XX  
 PS Claim 22; Page 64; 71pp; English.  
 XX  
 CC The present sequence is a recombinant Rana catesbeiana oocyte  
 CC ribonuclease (RacOR1) protein with Met22Leu Met57Leu. Carboxy terminal  
 CC end of recombinant RacOR1 has a covalently bound ligand binding moiety,  
 CC which can be a L12 antibody directed against CD22 on cancerous B cells  
 CC or human chorionic gonadotropin (hCG) effective against Kaposi's sarcoma  
 CC cells. Recombinant ribonucleases can be expressed in bacteria without an  
 CC N-terminal methionine due to the presence of a signal peptide that is  
 CC cleaved by bacteria. The soluble expression of ribonuclease allows the  
 CC proteins to be fused in-frame with ligand binding moieties to form  
 CC cytotoxic fusion proteins. They can be used for treatment of cancer and  
 CC autoimmune diseases.  
 CC  
 SQ Sequence 110 AA:  
 Query Match 98.2%; Score 594; DB 20; Length 110;  
 Best Local Similarity 99.1%; Pred. No. 2.1e-60;  
 Matches 109; Conservative 0; Mismatches 1; Indels 0; Gaps 0;  
 OY 1 QNMAFFQOKH IKPIIICNTILDNNIYIVGGCKRVNFTFISSATYKATCTGVINLNL 60  
 DB 1 QNMAFFQOKH IKPIIICNTILDNNIYIVGGCKRVNFTFISSATYKATCTGVINLNL 60  
 OY 61 STTRFOLNCTRTSITPRPCYSSRTETNYICVGCENQYPVHFGIGRCP 110  
 DB 61 STTRFOLNCTRTSITPRPCYSSRTETNYICVGCENQYPVHFGIGRCP 110  
 RESULT 2  
 ID AAY28876  
 AC AAY28876;  
 XX  
 DT 25-JAN-2000 (first entry)  
 XX  
 DE Recombinant Met(-1) RacOR1 Met22Leu Met57Leu-(His)6 protein.  
 XX  
 KW Met(-1) Rana catesbeiana ribonuclease Met22Leu Met57Leu-(His)6; RacOR1;  
 KW recombinant; CD22; covalently bound; L12 antibody; ligand binding moiety;  
 KW cancerous B cell; Kaposi's sarcoma; human chorionic gonadotropin; hCG;  
 KW signal peptide; recombinant ribonuclease; cytotoxic fusion protein;  
 KW cancer; bullfrog; RNase; autoimmune disease.  
 XX  
 OS Rana catesbeiana.  
 OS Synthetic.  
 XX  
 XX Key Location/Qualifiers  
 FT MISC-difference 1 /note= "(His)6 histidine tag attached to N-terminal Met"  
 FT MISC-difference 1 /note= "Met not found in wild type RacOR1"  
 FT MISC-difference 23 /note= "Wild type Met replaced with Leu"  
 FT MISC-difference 58 /note= "Wild type Met replaced with Leu"  
 FT MISC-difference 58 /note= "Wild type Met replaced with Leu"  
 XX  
 PN WO9950398-A2.  
 XX  
 XX 07-OCT-1999.  
 XX  
 XX 26-MAR-1999; 99WO-US06641.  
 PF

XX  
 PR 27-MAR-1998; 98US-0079751.  
 XX  
 PA (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
 XX  
 PI Newton DL, Rybak SM;  
 XX  
 DR MPI: 1999-610847/52.  
 DR N-PSDB: AA208133.  
 XX  
 PT New recombinant ribonucleases, used for killing target cells, e.g. for  
 PT treating cancers, viral infections or autoimmune diseases -  
 XX  
 PS Claim 22; Page 66; 71pp; English.  
 XX  
 CC The present sequence is a recombinant Rana catesbeiana oocyte  
 CC ribonuclease (RacOR1) protein with Met at position 1 attached to a  
 CC (His)6 tag, Met22Leu and Met58Leu. Carboxy terminal end of recombinant  
 CC RacOR1 has a covalently bound ligand binding moiety, which can be a L12  
 CC antibody directed against CD22 on cancerous B cells or human chorionic  
 CC gonadotropin (hCG) effective against Kaposi's sarcoma cells. Recombinant  
 CC ribonucleases can be expressed in bacteria without an N-terminal  
 CC methionine due to the presence of a signal peptide that is cleaved by  
 CC bacteria. The soluble expression of ribonuclease allows the proteins to  
 CC be fused in-frame with ligand binding moieties to form cytotoxic fusion  
 CC proteins. They can be used for treatment of cancer and autoimmune  
 CC diseases.  
 CC  
 SQ Sequence 111 AA:  
 Query Match 99.2%; Score 594; DB 20; Length 111;  
 Best Local Similarity 99.1%; Pred. No. 2.1e-60;  
 Matches 109; Conservative 0; Mismatches 1; Indels 0; Gaps 0;  
 OY 1 QNMAFFQOKH IKPIIICNTILDNNIYIVGGCKRVNFTFISSATYKATCTGVINLNL 60  
 DB 2 QNMAFFQOKH IKPIIICNTILDNNIYIVGGCKRVNFTFISSATYKATCTGVINLNL 61  
 OY 61 STTRFOLNCTRTSITPRPCYSSRTETNYICVGCENQYPVHFGIGRCP 110  
 DB 62 STTRFOLNCTRTSITPRPCYSSRTETNYICVGCENQYPVHFGIGRCP 111  
 RESULT 3  
 ID AAY28872  
 AC AAY28872;  
 XX  
 DT 25-JAN-2000 (first entry)  
 XX  
 DE Rana catesbeiana oocyte ribonuclease (RacOR1) amino acid sequence.  
 XX  
 KW Rana catesbeiana oocyte ribonuclease; RacOR1; covalently bound; CD22;  
 KW L12 antibody; ligand binding moiety; cancerous B cell; Kaposi's Sarcoma;  
 KW human chorionic gonadotropin; hCG; recombinant ribonuclease; bullfrog;  
 KW signal peptide; cytotoxic fusion protein; cancer; autoimmune disease;  
 KW RNase.  
 XX  
 OS Rana catesbeiana.  
 OS Synthetic.  
 XX  
 XX WO9950398-A2.  
 XX  
 XX 07-OCT-1999.  
 XX  
 XX 26-MAR-1999; 99WO-US06641.  
 XX  
 XX 27-MAR-1998; 98US-0079751.  
 XX  
 XX (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
 XX  
 XX Newton DL, Rybak SM;  
 PI

XX WPI: 1999-610847/52.  
DR N-PSDB; AAZ08130.  
PT New recombinant ribonucleases, used for killing target cells, e.g. for  
PS treating cancers, viral infections or autoimmune diseases  
XX Claim 22; Page 62; 71pp; English.  
XX The present sequence is a Rana catesbeiana oocyte ribonuclease (RacOR1)  
CC protein encoded by a cDNA modified for expression in E. coli. Carboxy  
CC terminal end of RacOR1 has a covalently bound ligand binding moiety,  
CC which can be a LL2 antibody directed against CD22 on cancerous B cells  
CC or human chorionic gonadotrophin (hCG) effective against Kaposi's  
CC sarcoma cells. Recombinant ribonucleases can be expressed in bacteria  
CC without an N-terminal methionine due to the presence of a signal peptide  
CC that is cleaved by bacteria. The soluble expression of ribonuclease  
CC allows the proteins to be fused in-frame with ligand binding moieties to  
CC form cytotoxic fusion proteins. They can be used for treatment of cancer  
CC and autoimmune diseases.  
XX Sequence 110 AA:  
SQ  
Query Match 98.5%; Score 590; DB 20; Length 110;  
Best Local Similarity 97.3%; Pred. No. 6.1e-60;  
Matches 107; Conservative 2; Mismatches 1; Indels 0; Gaps 0;  
QY 1 QNMAFTPOQKHIIKPIICNTILDNNIYVGGCKRVNFIISATYKAICTGVINLNL 60  
DB 1 QNMAFTPOQKHIIKPIICNTILDNNIYVGGCKRVNFIISATYKAICTGVINLNL 60  
QY 61 STTRFQNLNCTRTSITPRPCPYSSRTETNYICVGCENQYPVHFAGIGRCP 110  
DB 61 STTRFQNLNCTRTSITPRPCPYSSRTETNYICVGCENQYPVHFAGIGRCP 110  
RESULT 4  
AAZ28873 ID AAY28873 standard; Protein: 111 AA.  
XX AAY28873:  
AC 25-JAN-2000 (first entry)  
DT Recombinant Met(-1) RacOR1.  
DE Recombinant Met(-1) RacOR1.  
XX  
KM Recombinant Met(-1) Rana catesbeiana oocyte ribonuclease; RacOR1; CD22;  
KM covalently bound; LL2 antibody; ligand binding moiety; cancerous B cell;  
KM Kaposi's sarcoma; human chorionic gonadotrophin; hCG; signal peptide;  
KM recombinant ribonuclease; cytotoxic fusion protein; cancer; bullfrog;  
KM RNase; autoimmune disease.  
XX  
OS Rana catesbeiana.  
OS Synthetic.  
XX  
FH Key Location/Qualifiers  
FT Misc-difference 1 /note= "Met not found in wild type RacOR1"  
FT  
XX  
PN WO9950398-A2.  
PD 07-OCT-1999.  
XX  
PF 26-MAR-1999; 99WO-US06641.  
XX  
PR 27-MAR-1998; 98US-0079751.  
XX  
PA (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
XX  
PI Newton DL, Rybak SM;  
XX  
XX WPI: 1999-610847/52.  
DR N-PSDB; AAZ08131.  
PT

XX New recombinant ribonucleases, used for killing target cells, e.g. for  
PT treating cancers, viral infections or autoimmune diseases  
XX  
XX Claim 22; Page 63; 71pp; English.  
XX The present sequence is a recombinant Rana catesbeiana oocyte  
CC ribonuclease (RacOR1) protein with Met at position 1. Carboxy terminal  
CC end of recombinant RacOR1 has a covalently bound ligand binding moiety,  
CC which can be a LL2 antibody directed against CD22 on cancerous B cells or  
CC human chorionic gonadotrophin (hCG) effective against Kaposi's sarcoma  
CC cells. Recombinant ribonucleases can be expressed in bacteria without an  
CC N-terminal methionine due to the presence of a signal peptide that is  
CC cleaved by bacteria. The soluble expression of ribonuclease allows the  
CC proteins to be fused in-frame with ligand binding moieties to form  
CC cytotoxic fusion proteins. They can be used for treatment of cancer and  
CC autoimmune diseases.  
XX Sequence 111 AA:  
SQ  
Query Match 98.5%; Score 590; DB 20; Length 111;  
Best Local Similarity 97.3%; Pred. No. 6.2e-60;  
Matches 107; Conservative 2; Mismatches 1; Indels 0; Gaps 0;  
QY 1 QNMAFTPOQKHIIKPIICNTILDNNIYVGGCKRVNFIISATYKAICTGVINLNL 60  
DB 2 QNMAFTPOQKHIIKPIICNTILDNNIYVGGCKRVNFIISATYKAICTGVINLNL 61  
QY 61 STTRFQNLNCTRTSITPRPCPYSSRTETNYICVGCENQYPVHFAGIGRCP 110  
DB 62 STTRFQNLNCTRTSITPRPCPYSSRTETNYICVGCENQYPVHFAGIGRCP 111  
RESULT 5  
AAZ28877 ID AAY28877 standard; Protein: 110 AA.  
XX AAY28877:  
AC 25-JAN-2000 (first entry)  
DT Recombinant RacOR1 Gln1Ser amino acid sequence.  
DE Recombinant RacOR1 Gln1Ser amino acid sequence.  
XX  
KM Recombinant Rana catesbeiana oocyte ribonuclease; RacOR1 Gln1Ser; CD22;  
KM covalently bound; LL2 antibody; ligand binding moiety; cancerous B cell;  
KM bullfrog; Kaposi's sarcoma; human chorionic gonadotrophin; hCG; RNase;  
KM signal peptide; recombinant ribonuclease; cytotoxic fusion protein;  
KM cancer; autoimmune disease.  
XX  
OS Rana catesbeiana.  
OS Synthetic.  
XX  
FH Key Location/Qualifiers  
FT Misc-difference 1 /note= "Wild type Gln replaced with Ser"  
FT  
XX  
PN WO9950398-A2.  
PD 07-OCT-1999.  
XX  
PF 26-MAR-1999; 99WO-US06641.  
XX  
PR 27-MAR-1998; 98US-0079751.  
XX  
PA (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
XX  
PI Newton DL, Rybak SM;  
XX  
XX WPI: 1999-610847/52.  
DR N-PSDB; AAZ08134..  
PT New recombinant ribonucleases, used for killing target cells, e.g. for  
PT treating cancers, viral infections or autoimmune diseases

XX Claim 22; Page 67; 71pp; English.

XX  
 CC The present sequence is a recombinant Rana catesbeiana oocyte  
 CC ribonuclease (RacOR1) protein with Gln1Ser. Carboxy terminal end of  
 CC recombinant RacOR1 has a covalently bound ligand binding moiety, which  
 CC can be a IL2 antibody directed against CD22 on cancerous B cells or  
 CC human chorionic gonadotropin (hCG) effective against Kaposi's sarcoma  
 CC cells. Recombinant ribonucleases can be expressed in bacteria without an  
 CC N-terminal methionine due to the presence of a signal peptide that is  
 CC cleaved by bacteria. The soluble expression of ribonuclease allows the  
 CC proteins to be fused in-frame with ligand binding moieties to form  
 CC cytotoxic fusion proteins. They can be used for treatment of cancer and  
 CC autoimmune diseases.

SQ Sequence 110 AA:

Query Match 97.7%; Score 585; DB 20; Length 110;  
 Best Local Similarity 97.2%; Pred. No. 2.3e-59;  
 Matches 106; Conservative 2; Mismatches 1; Indels 0; Gaps 0;

OY 2 NMATFOQKHIIKTPICNTIIDNNIYIVGGCKRVNFTIISATVKAICTGVINLVLS 61  
 DB 2 NMATFOQKHIIINTPICNTIDNNIYIVGGCKRVNFTIISATVKAICTGVINLVLS 61  
 OY 62 TTRFQNLNCTRTSITPRCPYSSRTETNYICVKCENQYPVHFAGIGRCP 110  
 DB 62 TTRFQNLNCTRTSITPRCPYSSRTETNYICVKCENQYPVHFAGIGRCP 110

RESULT 6  
 AAY28878  
 ID AAY28878 standard; Protein: 111 AA.

XX  
 AC AAY28878;  
 AC  
 DT 25-JAN-2000 (first entry)  
 DT  
 DE Recombinant Met(-1) RacOR1 Gln1Ser amino acid sequence.

XX  
 KW Recombinant Met(-1) Rana catesbeiana oocyte ribonuclease Gln1Ser; RacOR1;  
 KW covalently bound; IL2 antibody; ligand binding moiety; cancerous B cell;  
 KW Kaposi's sarcoma; human chorionic gonadotropin; hCG; signal peptide;  
 KW recombinant ribonuclease; cytotoxic fusion protein; cancer; bullfrog;  
 KW CD22; RNase; autoimmune disease.

XX  
 OS Rana catesbeiana.  
 OS Synthetic.

XX  
 FH Key Location/Qualifiers  
 FT Misc-difference 1 /note= "Met not found in wild type RacOR1"  
 FT Misc-difference 2 /note= "Wild type Gln replaced with Ser"

XX  
 PN WO9950398-AZ.  
 PN  
 PD 07-OCT-1999.  
 PD  
 XX 26-MAR-1999; 99WO-US06641.  
 XX  
 PR 27-MAR-1998; 98US-0079751.  
 PR  
 XX (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
 XX  
 PA Newton DL, Rybak SM;  
 PI  
 PI  
 DR WPI: 1999-610847/52.  
 DR N-PSDB; AA208135.  
 DR  
 PT New recombinant ribonucleases, used for killing target cells, e.g. for  
 PT treating cancers, viral infections or autoimmune diseases  
 XX

PS Claim 22; Page 68; 71pp; English.

XX  
 CC The present sequence is a recombinant Rana catesbeiana ribonuclease  
 CC (RacOR1) protein with Met at position 1 and Gln2Ser. Carboxy terminal  
 CC end of recombinant RacOR1 has a covalently bound ligand binding moiety, which  
 CC can be a IL2 antibody directed against CD22 on cancerous B cells or human  
 CC chorionic gonadotropin (hCG) effective against Kaposi's sarcoma cells.  
 CC Recombinant ribonucleases can be expressed in bacteria without an N-  
 CC terminal methionine due to the presence of a signal peptide that is  
 CC cleaved by bacteria. The soluble expression of ribonuclease allows the  
 CC proteins to be fused in-frame with ligand binding moieties to form  
 CC cytotoxic fusion proteins. They can be used for treatment of cancer and  
 CC autoimmune diseases.

SQ Sequence 111 AA:

Query Match 97.7%; Score 585; DB 20; Length 111;  
 Best Local Similarity 97.2%; Pred. No. 2.3e-59;  
 Matches 106; Conservative 2; Mismatches 1; Indels 0; Gaps 0;

OY 2 NMATFOQKHIIKTPICNTIIDNNIYIVGGCKRVNFTIISATVKAICTGVINLVLS 61  
 DB 3 NMATFOQKHIIINTPICNTIDNNIYIVGGCKRVNFTIISATVKAICTGVINLVLS 62  
 OY 62 TTRFQNLNCTRTSITPRCPYSSRTETNYICVKCENQYPVHFAGIGRCP 110  
 DB 63 TTRFQNLNCTRTSITPRCPYSSRTETNYICVKCENQYPVHFAGIGRCP 111

RESULT 7  
 AAY33321  
 ID AAY33321 standard; Protein: 111 AA.

XX  
 AC AAY33321;  
 AC  
 DT 29-NOV-1999 (first entry)  
 DT  
 DE Frog lectin protein fragment.

XX  
 KW Cytotoxic; RNase; ribonuclease; pancreatic; antibody; light chain;  
 KW heavy chain; cell surface marker; treatment; tumor; viral infection;  
 KW parasite infection; immune dysfunctional cell; autoimmune disease;  
 KW contraceptive; cell separation; transplantation; bone marrow ablation;  
 KW leukemia cell; T-cell; graft-versus-host disease; bullfrog; lectin.

XX  
 OS Rana catesbeiana.  
 OS  
 PN US5955073-A.  
 PN  
 PD 21-SEP-1999.  
 PD  
 XX 09-JUL-1997; 97US-0891848.  
 XX  
 PR 22-SEP-1993; 93US-0125462.  
 PR 22-OCT-1991; 91US-0779195.  
 PR 20-APR-1990; 90US-0510696.  
 PR 04-FEB-1993; 93US-0014082.  
 PR  
 XX (USSH ) US DEPT HEALTH & HUMAN SERVICES.  
 XX  
 PA Rybak SM, Newton DL, Nicholls PJ, Youle RJ;  
 PI  
 PI  
 DR WPI: 1999-560488/47.  
 DR  
 XX Recombinantly fused pancreatic RNase-targeting proteins useful for  
 XX treating tumors, infections, immune or autoimmune disorders and as a  
 XX contraceptive  
 XX  
 PS Example 3; Fig 19; 47pp; English.

CC This invention describes a novel nucleic acid construct comprising  
 CC sequences encoding functional pancreatic RNase and a second protein  
 CC (preferably the light and heavy chains of an antibody) which binds a



Query Match 45.7%, Score 273.5; DB 20; Length 104;  
Best Local Similarity 48.6%, Pred. No. 1.2e-23;  
Matches 54; Conservative 15; Mismatches 33; Indels 9; Gaps 4;

1 QMATTFOOKHIIKT-PIICNTILDNNIYIVGGCKRVNTFISSATTYKATCGVI-NLN 58  
1 QDWLTFOKKHILTNTRDVCNNIMSTNLF---HCKDKNTFTYSRPEPVKAICKGIASKN 56

59 VLSSTRFQLMTCTRTSTIPRCPYSSRTEWYICVKCENQYPVHFAIGRC 109  
57 VLTTSERYLSDC---NVTSRPCKYKIKKSTNTFCVTCENQAPVHFGVGHG 104

## RESULT 10

AAV28869 standard; Protein: 105 AA.

AAV28869;

25-JAN-2000 (first entry)

Recombinant Met(-1) RapLRI Met23Leu-(His)6 protein.

Rana pipiens liver ribonuclease Met23Leu-(His)6; RapLRI;  
CD22; covalently bound; IL2 antibody; ligand binding moiety; RNase;  
cancerous B cell; Kaposi's sarcoma; human chorionic gonadotropin; hCG;  
signal peptide; recombinant ribonuclease; cytotoxic fusion protein;  
cancer; frog; autoimmune disease.

Rana pipiens.  
Synthetic.

Location/Qualifiers

Key

Misc-difference 1 /note= "(His)6 histidine tag attached to N-terminal Met"

Misc-difference 24 /note= "Met not found in wild type RapLRI"

Misc-difference 24 /note= "Wild type Met replaced with Leu"

WO950398-A2.

07-OCT-1999.

26-MAR-1999; 99WO-US06641.

27-MAR-1998; 98US-0079751.

(USSH ) US DEPT HEALTH &amp; HUMAN SERVICES.

Newton DL, Rybak SM;

WPI: 1999-610847/52.

N-PSDB; AA208127.

New recombinant ribonucleases, used for killing target cells, e.g. for  
treating cancers, viral infections or autoimmune diseases

Claim 4; Page 59; 71pp; English.

The present sequence is a recombinant Rana pipiens ribonuclease protein  
(RapLRI) with Met at position 1 attached to (His)6 tag and Met24Leu.  
Carboxy terminal end of recombinant RapLRI has a covalently bound ligand  
binding moiety, which can be a IL2 antibody directed against CD22 on  
cancerous B cells or human chorionic gonadotropin (hCG) effective  
against Kaposi's sarcoma cells. Recombinant ribonucleases can be  
expressed in bacteria without an N-terminal methionine due to the  
presence of a signal peptide that is cleaved by bacteria. The soluble  
expression of ribonuclease allows the proteins to be fused in-frame with  
ligand binding moieties to form cytotoxic fusion proteins. They can be  
used for treatment of cancer and autoimmune diseases.

Sequence 105 AA;

Query Match 45.7%, Score 273.5; DB 20; Length 105;  
Best Local Similarity 48.6%, Pred. No. 1.2e-23;  
Matches 54; Conservative 15; Mismatches 33; Indels 9; Gaps 4;

1 QMATTFOOKHIIKT-PIICNTILDNNIYIVGGCKRVNTFISSATTYKATCGVI-NLN 58  
2 QDWLTFOKKHILTNTRDVCNNIMSTNLF---HCKDKNTFTYSRPEPVKAICKGIASKN 57

59 VLSSTRFQLMTCTRTSTIPRCPYSSRTEWYICVKCENQYPVHFAIGRC 109  
58 VLTTSERYLSDC---NVTSRPCKYKIKKSTNTFCVTCENQAPVHFGVGHG 105

## RESULT 11

AAV28865 standard; Protein: 104 AA.

AAV28865;

25-JAN-2000 (first entry)

Rana pipiens liver ribonuclease (RapLRI).

Rana pipiens liver ribonuclease; RapLRI; covalently bound; IL2 antibody;  
ligand binding moiety; CD22; cancerous B cell; Kaposi's Sarcoma; frog;  
human chorionic gonadotropin; hCG; recombinant ribonuclease; RNase;  
signal peptide; cytotoxic fusion protein; cancer; autoimmune disease.

Rana pipiens.

WO950398-A2.

07-OCT-1999.

26-MAR-1999; 99WO-US06641.

27-MAR-1998; 98US-0079751.

(USSH ) US DEPT HEALTH &amp; HUMAN SERVICES.

Newton DL, Rybak SM;

WPI: 1999-610847/52.

N-PSDB; AA208124.

New recombinant ribonucleases, used for killing target cells, e.g. for  
treating cancers, viral infections or autoimmune diseases

Claim 1; Page 55; 71pp; English.

The present sequence is Rana pipiens liver ribonuclease (RapLRI)  
protein. Carboxy terminal end of RapLRI has a covalently bound  
ligand binding moiety, which can be a IL2 antibody directed against  
CD22 on cancerous B cells or human chorionic gonadotropin (hCG).  
effective against Kaposi's Sarcoma cells. Recombinant ribonucleases can  
be expressed in bacteria without an N-terminal methionine due to the  
presence of a signal peptide that is cleaved by bacteria. The soluble  
expression of ribonuclease allows the proteins to be fused in-frame with  
ligand binding moieties to form cytotoxic fusion proteins. They can be  
used for treatment of cancer and autoimmune diseases.

Sequence 104 AA;

Query Match 45.3%, Score 271.5; DB 20; Length 104;  
Best Local Similarity 47.7%, Pred. No. 2e-23;  
Matches 53; Conservative 16; Mismatches 33; Indels 9; Gaps 4;

1 QMATTFOOKHIIKT-PIICNTILDNNIYIVGGCKRVNTFISSATTYKATCGVI-NLN 58  
1 QDWLTFOKKHILTNTRDVCNNIMSTNLF---HCKDKNTFTYSRPEPVKAICKGIASKN 56

59 VLSSTRFQLMTCTRTSTIPRCPYSSRTEWYICVKCENQYPVHFAIGRC 109

Db 57 VLTSEFYLSDC---NVTSPCKYKLRKSTNFCVTCENQAPVHFVGVGHC 104

## RESULT 12

AAV28867 ID AAY28867 standard; Protein: 105 AA.

AC AAY28867;

DT 25-JAN-2000 (first entry)

DE Recombinant Met(-1) RapLRL.

XX Recombinant Met(-1) Rana pipiens ribonuclease; RapLRL; CD22; RNase;

KM covalently bound; LL2 antibody; ligand binding moiety; cancerous B cell;

KM Kaposi's sarcoma; human chorionic gonadotropin; hCG; signal peptide;

KM recombinant ribonuclease; cytotoxic fusion protein; cancer; frog;

XX autoimmune disease.

XX Rana pipiens.

OS Synthetic.

XX Key

FT Misc-difference 1

FT /note= "Met not found in wild type RapLRL"

XX W09950398-A2.

PD 07-OCT-1999.

XX 26-MAR-1999; 99WO-US06641.

PF 27-MAR-1998; 98US-0079751.

XX (USSH ) US DEPT HEALTH & HUMAN SERVICES.

XX Newton DL, Rybak SM;

XX WPI: 1999-610347/52.

DR N-PSDB; AA208126.

XX New recombinant ribonucleases, used for killing target cells, e.g. for

XX treating cancers, viral infections or autoimmune diseases

XX Claim 34; Page 57; 71pp; English.

XX The present sequence is a recombinant Rana pipiens ribonuclease (RapLRL)

CC protein with Met at position 1. Carboxy terminal end of recombinant

CC RapLRL has a covalently bound ligand binding moiety, which can be a LL2

CC antibody directed against CD22 on cancerous B cells or human chorionic

CC gonadotropin (hCG) effective against Kaposi's sarcoma cells. Recombinant

CC ribonucleases can be expressed in bacteria without an N-terminal

CC methionine due to the presence of a signal peptide that is cleaved by

CC bacteria. The soluble expression of ribonuclease allows the proteins to

CC be fused in-frame with ligand binding moieties to form cytotoxic fusion

CC proteins. They can be used for treatment of cancer and autoimmune

CC diseases.

XX Sequence 105 AA;

XX Query Match 45.3%; Score 271.5; DB 20; Length 105;

Best Local Similarity 47.7%; Pred. No. 2, 1e-23;

Matches 53; Conservative 16; Mismatches 33; Indels 9; Gaps 4;

QY 1 QMNAFQOQHIIKT-PIICNTILDNNIYVGGCKRVNFTIISATVKAICTGVI-NLN 58

DB 2 QMNAFQOQHIIKT-PIICNTILDNNIYVGGCKRVNFTIISATVKAICTGVI-NLN 57

QY 59 VLTSEFYLSDC---NVTSPCKYKLRKSTNFCVTCENQAPVHFVGVGHC 109

DB 58 VLTSEFYLSDC---NVTSPCKYKLRKSTNFCVTCENQAPVHFVGVGHC 105

## RESULT 13

AAV28879 ID AAY28879 standard; Protein: 127 AA.

AC AAY28879;

DT 25-JAN-2000 (first entry)

DE Rana pipiens Clone 5a1b ribonuclease.

XX Rana pipiens ribonuclease Clone 5a1b; RapLRL; covalently bound; RNase;

KM LL2 antibody; ligand binding moiety; CD22; cancerous B cell; onconase;

KM Kaposi's Sarcoma; human chorionic gonadotropin; hCG; cancer;

KM recombinant ribonuclease; frog; signal peptide; cytotoxic fusion protein;

XX autoimmune disease.

XX Rana pipiens.

OS Key

FT Peptide

FT /label= Signal-peptide

FT /note= "Putative"

FT Protein

XX W09950398-A2.

PD 07-OCT-1999.

XX 26-MAR-1999; 99WO-US06641.

PF 27-MAR-1998; 98US-0079751.

XX (USSH ) US DEPT HEALTH & HUMAN SERVICES.

XX Newton DL, Rybak SM;

XX WPI: 1999-610847/52.

DR N-PSDB; AA208136.

XX New recombinant ribonucleases, used for killing target cells, e.g. for

XX treating cancers, viral infections or autoimmune diseases

XX Disclosure; Page 69; 71pp; English.

XX The present sequence is a Rana pipiens Clone 5a1b ribonuclease (RapLRL).

CC It is encoded by Clone 5a1b cDNA obtained from Rana pipiens liver mRNA

CC library. It exhibits differences with Onconase (RTM) at amino acid

CC residues 11, 20, 85 and 103. Carboxy terminal end of RapLRL has a

CC covalently bound ligand binding moiety, which can be a LL2 antibody

CC directed against CD22 on cancerous B cells or human chorionic

CC gonadotropin (hCG) effective against Kaposi's Sarcoma cells. Recombinant

CC ribonucleases can be expressed in bacteria without an N-terminal

CC methionine due to the presence of a signal peptide that is cleaved by

CC bacteria. The soluble expression of ribonuclease allows the proteins to

CC be fused in-frame with ligand binding moieties to form cytotoxic fusion

CC proteins. They can be used for treatment of cancer and autoimmune

CC diseases.

XX Sequence 127 AA;

XX Query Match 45.3%; Score 271.5; DB 20; Length 127;

Best Local Similarity 47.7%; Pred. No. 2, 6e-23;

Matches 53; Conservative 16; Mismatches 33; Indels 9; Gaps 4;

QY 1 QMNAFQOQHIIKT-PIICNTILDNNIYVGGCKRVNFTIISATVKAICTGVI-NLN 58

DB 24 QMNAFQOQHIIKT-PIICNTILDNNIYVGGCKRVNFTIISATVKAICTGVI-NLN 79

QY 59 VLTSEFYLSDC---NVTSPCKYKLRKSTNFCVTCENQAPVHFVGVGHC 109

DB 80 VLTSEFYLSDC---NVTSPCKYKLRKSTNFCVTCENQAPVHFVGVGHC 127

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RESULT 14
AAM30301
ID AAM30301 standard; protein; 104 AA.
XX
AC AAM30301;
XX
DT 09-JUN-1998 (first entry)
XX
DE Recombinant onc protein.
XX
KW Onc; oncanase; ribonuclease; frog; antitumour; pancreatic cancer;
KW human immunodeficiency virus type-1; HIV1; replication.
XX
OS Rana pipiens.
XX
PN W09738112-A1.
XX
PD 16-OCT-1997.
XX
PF 04-APR-1997; 97MO-US05675.
XX
PR 04-APR-1996; 96US-0626288.
XX
PA (USSH ) US DEPT HEALTH & HUMAN SERVICES.
XX
PI Ardeit W, Boix E, Vasandani VM, Wu YN, Youle RJ;
XX
DR WPI; 1997-512725/47.
XX
PT Recombinant Onc protein with glutamine residue at position 1 -
PT useful as antitumour and antiviral agent, also as cell culture
PT selection agent
XX
PS Claim 1; Page 28; 35pp; English.
XX
CC This sequence represents a recombinant Onc protein comprising a 104 amino
CC acid sequence having Gln at position 1. Onc, a ribonuclease from Rana
CC pipiens oocytes, is known as an antitumour agent (e.g. for treating
CC pancreatic cancer) and inhibitor of human immunodeficiency virus type-1
CC replication. It can be used therapeutically or as a cell-culture
CC selection agent, e.g. to identify gene therapy compositions able to
CC inhibit tumour growth.
XX
SQ Sequence 104 AA;
Query Match 44.7%; Score 267.5; DB 18; Length 104;
Best Local Similarity 47.7%; Pred. No. 5.9e-23;
Matches 53; Conservative 16; Mismatches 33; Indels 9; Gaps 4;
QY 1 QNMATFOOKHIKT-PIICNTILDNNIYVGGCKRVNPFITISSATYKAICTGYI-NLN 58
DB 1 QDMLEFQKKHINTNDVDCDNIIMSTNLF---HCKDKNTFYISRPVPKAIKGIASKN 56
QY 59 VLSITRFQNLNCTRTSITPRPCPYSSRTETNYICVKCENQYVHPAGIGRC 109
DB 57 VLTTSSEFYLSDC---NVTSPCKYKLLKSKSTNKFVTCENQAPVHFGVGVGSC 104

RESULT 15
AAB31666
ID AAB31666 standard; protein; 104 AA.
XX
AC AAB31666;
XX
DT 30-APR-2001 (first entry)
XX
DE Amino acid sequence of a frog ribonuclease protein.
XX
KW Frog; ribonuclease; ranpirinase; RNase.
XX
OS Rana pipiens.

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XX
FH Key Location/Qualifiers
FT Modified-site 1
FT FT /note= "this Gln is autocyclised to pyroglutamic acid"
XX
PN US6175003-B1.
XX
PD 16-JAN-2001.
XX
PF 10-SEP-1999; 99US-0394268.
XX
PR 10-SEP-1999; 99US-0394268.
XX
PA (ALFA-) ALFACELL CORP.
XX
PI Saxena SK;
XX
DR WPI; 2001-167808/17.
XX
PT New nucleic acids encoding a ribonuclease (Rnase), useful for the
XX precise targeting of Rnase to a predetermined cell receptor -
XX
PS Claim 1; Columns 5-6; 7pp; English.
XX
CC The present sequence represents a frog ribonuclease protein (ranpirinase)
CC (RNase). The specification describes a synthetic ribonuclease protein,
CC in which the addition of cysteine in the ribonuclease facilitates the
CC chemical linking of a targeting molecule by the single reactive
CC sulfhydryl group. The specification also describes a method for the
CC production of ranpirinase using DNA technology instead of processing
CC biological material. The re-engineering of the protein molecule allows
CC easier attachment to a targeting molecule thereby making it possible for
CC the ribonuclease to be delivered to a particular cell receptor where it
CC might be most effective.
XX
SQ Sequence 104 AA;
Query Match 44.7%; Score 267.5; DB 22; Length 104;
Best Local Similarity 47.7%; Pred. No. 5.9e-23;
Matches 53; Conservative 16; Mismatches 33; Indels 9; Gaps 4;
QY 1 QNMATFOOKHIKT-PIICNTILDNNIYVGGCKRVNPFITISSATYKAICTGYI-NLN 58
DB 1 QDMLEFQKKHINTNDVDCDNIIMSTNLF---HCKDKNTFYISRPVPKAIKGIASKN 56
QY 59 VLSITRFQNLNCTRTSITPRPCPYSSRTETNYICVKCENQYVHPAGIGRC 109
DB 57 VLTTSSEFYLSDC---NVTSPCKYKLLKSKSTNKFVTCENQAPVHFGVGVGSC 104

Search completed: June 25, 2003, 14:48:40
Job time : 33 secs

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